



Introduction

It is generally accepted that fats and oils play a major role in human nutrition as an important dietary source of energy and supply the body with essential fatty acids. Even though the Western diet is relatively rich in fat, the majority of the population does not consume adequate levels of the important ω -3 long chain PUFA, naturally present in marine foods among others.

DHA belong to the group of the important ω -3 long chain PUFA and research concerning this class of compounds have been going on since 1929 (Burr and Burr). The majority of the studies reported in the literature suggest that the decline in DHA intake could have serious implications for public health. Several clinical studies have shown that DHA supplementation resulted in lowering of serum triglyceride levels and improving the HDL:LDL cholesterol ratio. These findings suggest that supplementing with DHA might be cardioprotective. Other studies indicate that certain plasma DHA levels are important in regard to infant development. Prevention and treatment of other diseases by supplementation of DHA has been and still are under investigation such as: cancer, inflammatory diseases, psoriasis and age related diseases. This list is only intended as examples of studies reported in the literature and should not be considered as all inclusive.

By fortification of common foods the intake of certain basic nutrients such as DHA could be increased and people could then live healthier without changing their dietary habits. This is the basic for the growing interest for functional foods. Functional food can be defined as foods containing ingredients that provide health benefits or even disease prevention.

It is no news that health margarine such as polyunsaturated margarine can be produced since these products have been on the market for decades. These products contain a declared minimum level of polyunsaturated and essential fatty acids, and normally low amounts of salt. Additionally, spreadable fat products containing fish oils and thus high in particularly ω -3 long chain fatty acids have been on the market for years in Europe, manufactured under the names „Pact“, „Omega“ and „Blue Gaio“.

Experimental work

The source of DHA in the defined margarine does not originate from fish oil like the above listed products, but from a marine microalgae strain that produce vegetable oil containing large amounts of DHA, i.e. 40% DHA content of the vegetable oil. The defined fat blend used in this study consisted additionally of interesterified fat in order to minimise the *trans* fatty acid content in the margarine. Various degrees of cooling along with different degrees of mechanical sheer, i.e. RPM of pin rotor machine were applied. In addition, it was examined if the margarine could achieve a 6 months shelf life, based on analytical results and organoleptic evaluation.



GS Pin rotor machine

The interest in producing margarine and spreads based on interesterified fat blends have arose from the increasing awareness of the influence of *trans* fatty acids on health. Traditionally, margarine contains partially hydrogenated vegetable oil since hydrogenated fats crystallise relatively faster than unhydrogenated fats and provide the finished margarine with the necessary consistency. But during the hydrogenation process various *trans* fatty acids are formed and at present these isomers are thought to be nutritionally undesirable. Several studies reported in the last decade seem to indicate a correlation between a certain intake of these isomers and the risk of cardiovascular heart disease.

On this basis, there seems to be a global trend towards a reduction of the *trans* fatty acid content in margarine and other foods especially after Denmark by law has banned *trans* fatty acid and US FDA recommends reducing the contents to less than 1% of the energy. Consequently, the Food Industry will be forced to decrease the use of partially hydrogenated fat substantially, leading to a need of a replacement that provides among other things the margarine and spreads with the required functionalities and characteristics.

Table 1 shows the composition of the fat blend used for the production of the soft table margarine containing DHA. The hard stock of the defined fat blend consisted mainly of interesterified oil in order to minimise the *trans* fatty acid content in the product. The interesterified fat blend, Body fat 62, was supplied by AarhusKarlshamn, Denmark, and chosen since Body fat 62 is applicable for various margarines and spreads. Martek Biosciences Inc., USA provided vegetable oil from microalgae containing 40% DHA. The antioxidants, ascorbyl palmitate and tocopherols are added to the oil by the manufacturer.

Ingredients	DHA-blend (%)	Typical (%)
Body fat 62	37	-
Hydrogenated vegetable oil 41°/42°C	-	20
Hydrogenated vegetable oil 35°/36°C	-	20
Liquid oil	62	60
DHA	1	-

Table 1: Composition of the fat blend

TRANS FREE TABLE MARGARINE CONTAINING DHA

The emulsifier and flavour were supplied by Danisco A/S, Denmark. Apart from lecithin being an emulsifier, lecithin and ascorbyl palmitate were additionally added as antioxidants to the margarine fat blend in order to achieve extended oxidative stability. Studies have shown that binary systems of ascorbyl palmitate and lecithin are strongly synergistic in delaying peroxidation. However, the studies also showed that ternary blends of tocopherol (2%), ascorbyl-palmitate (0.1%) and lecithin (0.5%) provide the greatest protection against autooxidation.

A typical soft table margarine fat blend is additionally listed for comparison. It can be noted that this blend contains partially hydrogenated soybean oil, thus exhibits a content of *trans* fatty acids of approximately 15-20% depending on the hydrogenation process.

Prior to margarine production, information on the characteristics of the fat blend was obtained. The Solid Fat Content (SFC) profile was determined and the values shown correspond to SFC values normally obtained for a typical soft table margarine.

Temperature	DHA-blend (%)	Typical (%)
10°C	27	28
20°C	14	14
30°C	5	3
40°C	-	-

Table 2: Solid Fat Content (SFC) values

Low *trans* and zero *trans* margarine can be prepared from interesterified fat blends. Going through some of the studies reported in the literature, it is evident that interesterified fat blends show different crystallisation behaviour than hydrogenated fat blends. Therefore, attention should be paid to the composition and to the processing parameters when interesterified fat blends are involved in margarine and spreads production. However, quality margarine and spreads can be achieved exhibiting excellent sensoric properties.

The soft table margarine was produced based on the following set-up:

HPP → first chiller with intermediate crystalliser → second chiller → third chiller → pin rotor machine (volume 2 liters) → filling machine. The emulsion is transferred through the HPP with a capacity of 90 kg/hour to the first cooling section of the SSHE, the Perfector. Here sub cooling of the emulsion, nucleation and subsequently crystal growth takes place. Before entering the second cooling section the semi crystallised emulsion is kneaded in the intermediate crystalliser mounted directly on the shaft on the first cooling tube. Intermediate crystalliser is employed in order to yield mechanical work to the product and hereby ensure homogeneity, plasticity and spreadability. After the second and the third cooling tube the crystallised fat is finally kneaded in the pin rotor machine before filling.



GS Perfector 125

Where the rotation, i.e. the RPM of the pin in the intermediate crystalliser depends on the rotation of the rotor shaft in the chilling tube, the pin rotor machine is an independent machine with variable speed of the pin rotor. In these trials low, medium and high RPM were applied. The following table 3 shows the processing conditions.

Sample	Cooling medium temperature (NH ₃)			Pin Rotor
	1 st chiller	2 nd chiller	3 rd chiller	RPM
1	-5°C	-5°C	-5°C	50
2	-5°C	-5°C	-5°C	250
3	-5°C	-5°C	-5°C	500
4	-15°C	-5°C	-5°C	500
5	-15°C	-5°C	-5°C	250
6	-15°C	-5°C	-5°C	50

Table 3: Processing conditions

The trials can be grouped into two groups, sample 1-3 where the amount of cooling were for all 3 chilling tubes -5°C, and sample 4-6 where the amount of cooling in the first chilling tube was changed to -15°C. The cooling is applied by means of ammonia. In addition, the different RPM of the pin rotor machine were then applied for each cooling parameters.

The crystallisation process was followed during the processing of margarine by taking samples after each chilling tube and working unit for instant SFC determination at 20°C. This graph illustrates the differences in the instant SFC measured at various places in the processing of soft table margarine. The measurement SFC1 corresponds to the solid fat content after the first chilling tube, SFC2 correspond to the solid fat content after the second chilling tube, and so forth. The sample numbers correspond to the various processing parameters.

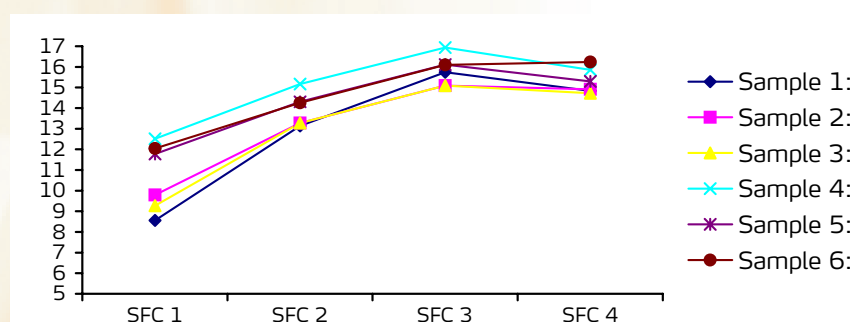


Figure 1: Instant SFC measured during the margarine processing.

It was examined if an optimal cooling temperature and/or RPM of the pin rotor machine could be determined by evaluating the various instant SFC profiles corresponding to the degrees of cooling and RPM of pin rotor machine.

The instant SFC profile of the all the samples show a steady increase in the SFC values measured after each cooling tube during processing. However, it can be noted sample 4-6 initially show a significantly higher content of solids at SFC1 and SFC2 when compared to sample 1-3, which not surprisingly means that intensive cooling onset the crystallisation process.

A stagnation or decrease in instant SFC value measured after the pin rotor machine may be expected. Since the pin worker functions as a kneading unit, the addition of mechanical work without cooling creates a stagnation for sample 2 and 6, and a decrease in the crystallisation for the rest of the samples. These results might indicate that optimal cooling temperature and RPM of the pin worker machine for the defined blend and processing parameters correspond to the parameters of sample 6. Sample 6 shows the highest value in SFC by the end of processing and additionally no decrease in SFC over the pin worker. The stagnation or even increase over the pin worker means that the solids created prior in the process is not melted due to too intense mechanical treatment.

Water droplet size and distribution was analysed by Nuclear Magnetic Resonance, NMR, using Minispec from Bruker, Germany. The most important parameter in characterising any emulsion is the size distribution. Two emulsions may have the same average droplet size and yet exhibit quite dissimilar behavior because of differences in their distribution of diameters.

Stability of the emulsion is one of the phenomena influenced by both relative size and size distribution. Size is an individual droplet property, but any property of one droplet is not an interesting value in the overall emulsion evaluation. The real interest is the entire size distribution of the emulsion.

	1	2	3	4	5	6
2.5 % <	0.9	0.9	1.0	1.2	1.2	1.3
50.0 % <	2.9	3.0	2.9	2.8	2.8	2.8
97.5 % <	9.6	9.6	9.1	6.8	6.4	5.7

Table 4: Water droplet size distribution

For all the samples it can be noted that less than 2.5% of the water droplets have a size of approximately 1 micron. No major differences were found for the mean value size distribution, since 50% of the droplets have a size of approximately 3 microns. However, the values for the remaining 2.5% of the water droplets show variation. The water droplets appear to be larger, i.e. 9.5 microns, for the samples produced with less intensive chilling when compared to sizes of around 6.4 microns for samples produced with more intensive chilling in the first cooling section of the Perfector.

Typical water droplet distribution values for margarine are found in to be in the range described above. However, in regard to low fat spreads the distribution might be much wider depending on the composition of the product. In addition, the results of the water droplet determination indicate that the RPM of the pin rotor machine does not significantly affect the water droplet size in soft table margarine.

Peroxide values of the produced margarine were measured by Martek Biosciences Corp. Using standardised AOCS methods (Cd8-53). In general, peroxide values of less than 5 will be accepted.

	1	2	3	4	5	6
3 months	1.19	1.16	1.17	1.30	1.26	1.25
6 months	2.57	2.20	2.29	2.17	2.23	2.03

Table 5: Peroxide values

It can be noted that none of the listed values are higher than 2.5, even after 6 months. No significant difference between the values for 3 and 6 months individually was found. However, significance was found between the values for 3 and 6 months ($P < 0.05$, two sample t-test, one sided). Since the peroxide value test alone is not a satisfactory measure of oxidation state due to potential side reactions, organoleptic evaluations were done. No overall differences in taste between the samples were found.

When comparing the subjective evaluation for this defined study, i.e. the organoleptic evaluation, and the objective measurements such as water droplet distribution and peroxide values, it was expected that greater differences were found. However, one has to remember that these results are indications, but due to the sample size no statistically significance can be determined.

Conclusion

It can be concluded that good quality soft table margarine containing DHA can be produced. The results obtained by droplet size determination suggest that the distribution is affected by the intensity of cooling and not by the RPM of the pin rotor. In order to establish statistically significant correlation between the objective water droplet size distribution method, and the subjective evaluation, further work needs to be done.

Work is still being done in order to clarify the differences in the crystallisation process when margarine is composed of interesterified fats. By optimising not only the composition of the interesterified fat blends, but also the processing parameters on the basis of analytical and processing results, a better understanding of the crystallisation process when interesterified fat blends are involved might be apparent.

Please contact GS for further information.

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